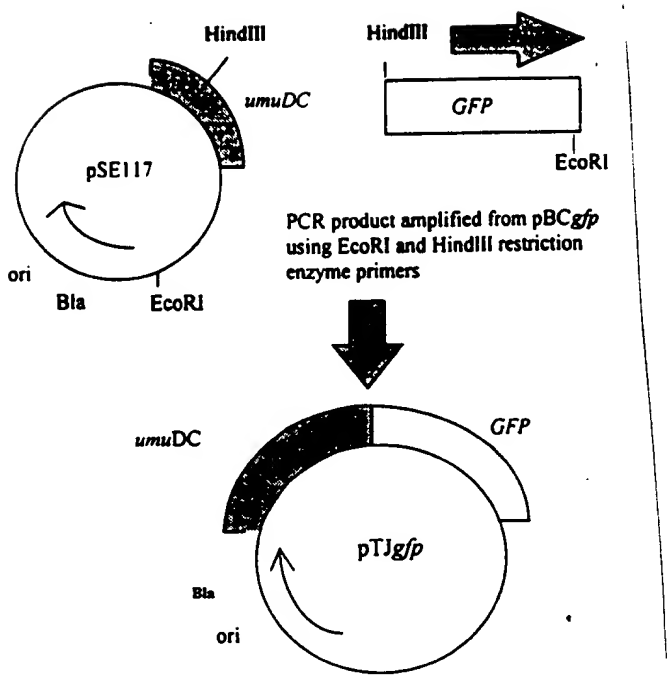

HindIII
HindIII
umuDC
GFP
pSE117
EcoRI
PCR product amplified from pBC*gfp* using EcoRI and HindIII restriction enzyme primers
ori
Bla
EcoRI
umuDC
GFP
pTJ*gfp*
Bla
ori

Fig 1

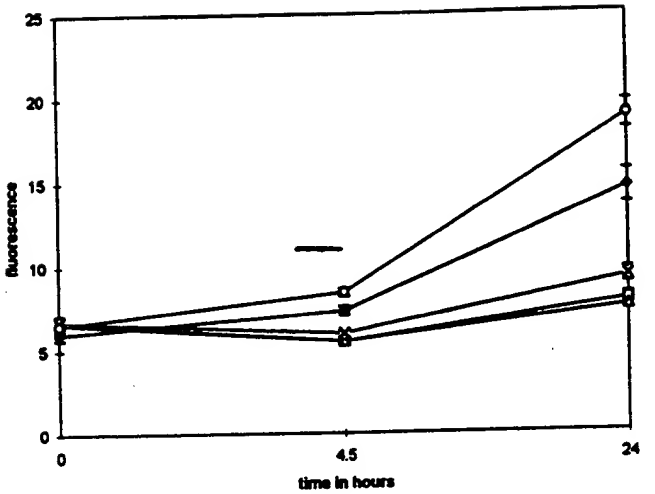

25
20
15
10
5
0
fluorescence
0
4.5
24
time in hours

Fig 2a

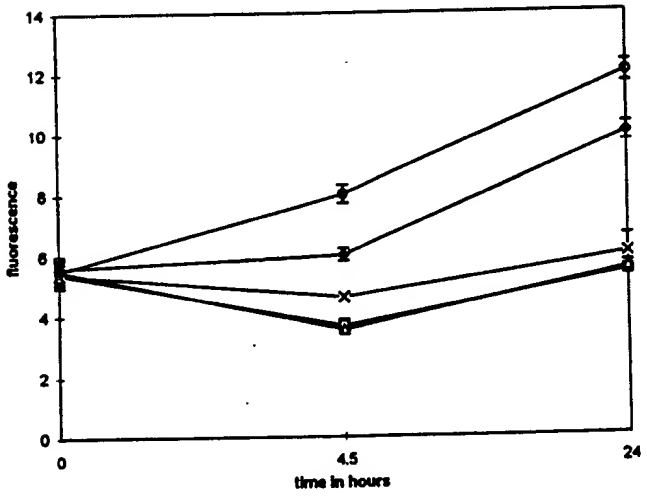

14
12
10
8
6
4
2
0
fluorescence
0
4.5
24
time in hours

Fig 2b

Fig 3a

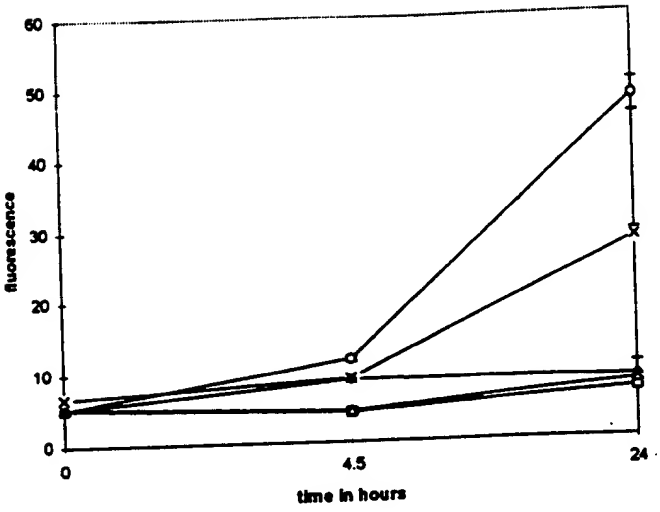
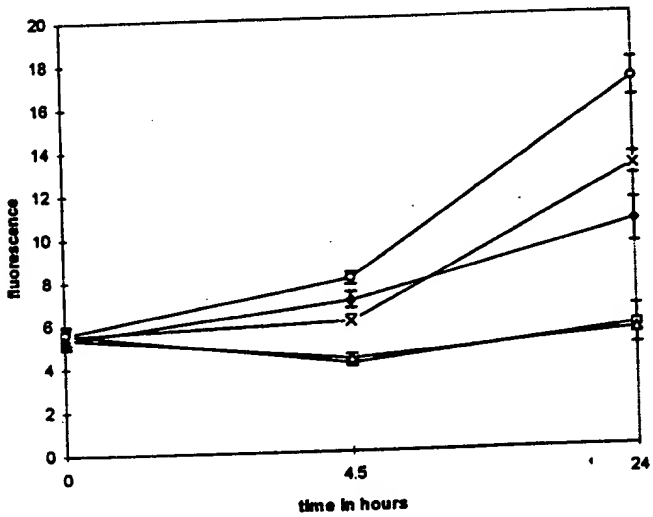

60
50
40
30
20
10
0
fluorescence
0
4.5
24
time in hours

Fig 3b


20
18
16
14
12
10
8
6
4
2
0
fluorescence
0
4.5
24
time in hours

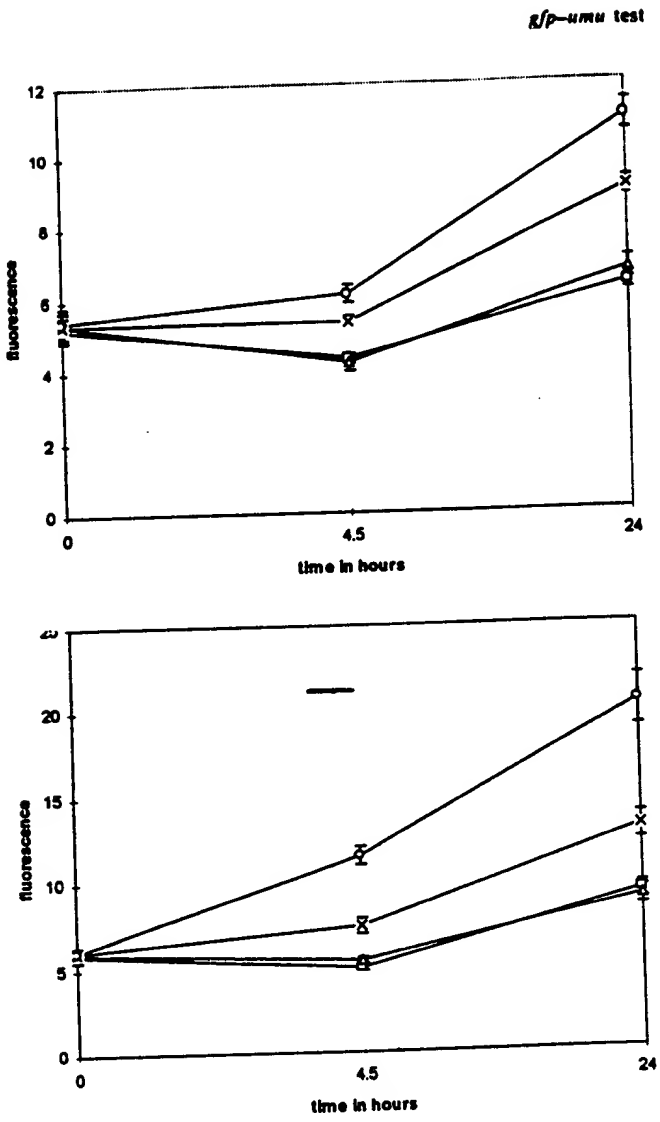

gfp-umu test
fluorescence
time in hours
0
4.5
24

Fig 4a

fluorescence
time in hours
0
4.5
24

Fig 4b

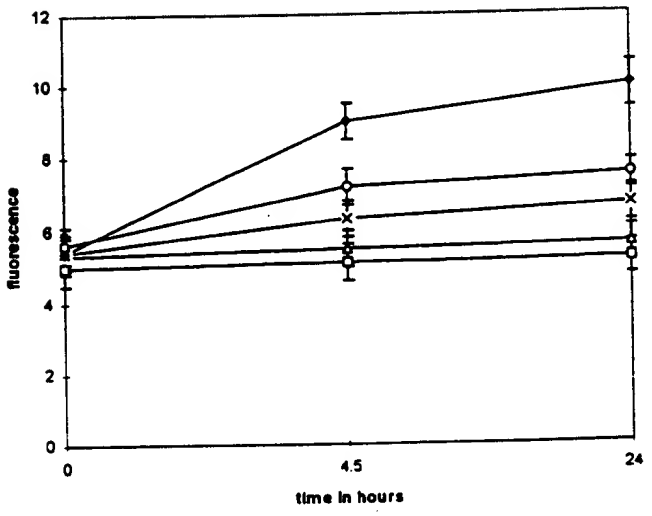


Fig 5

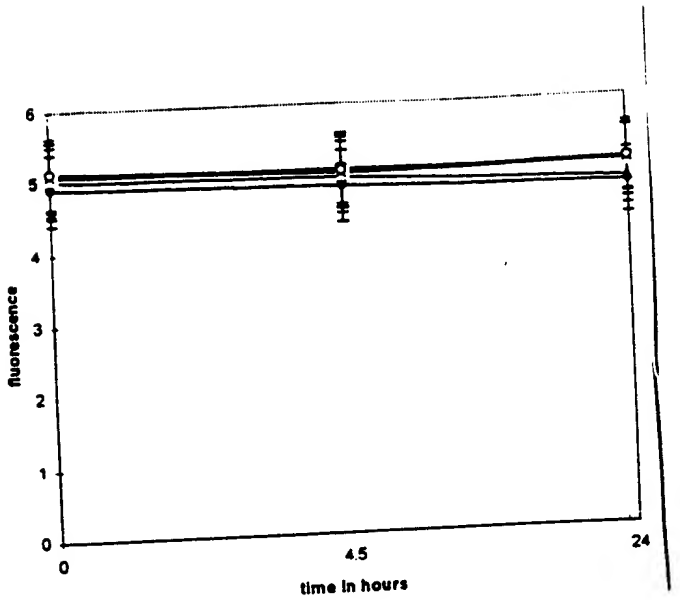


Fig 6

Figure 7 Histogram of mutant colony numbers from 40 parallel cultures following exposure to MNNG

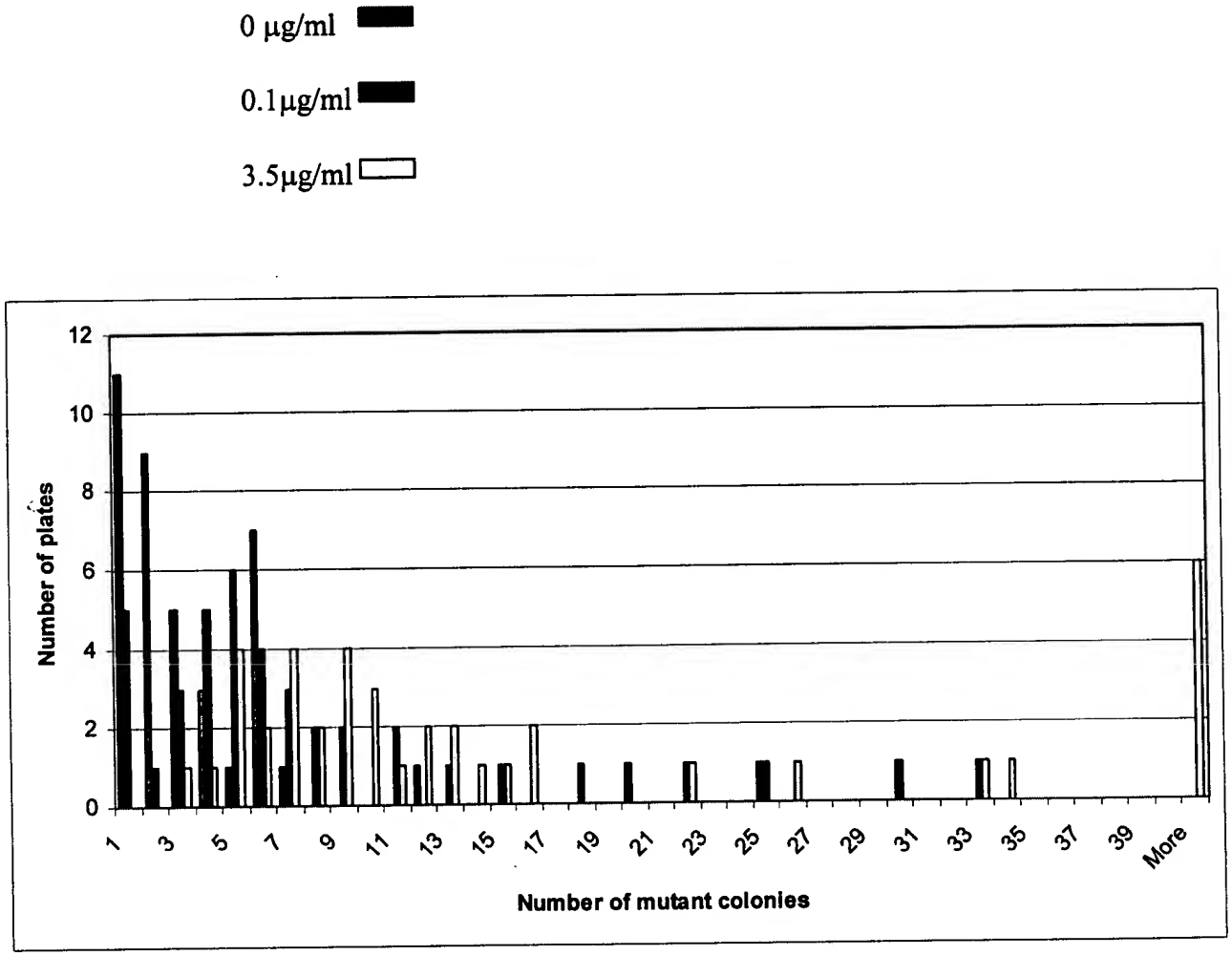


Figure 8 Histogram of mutant colony numbers from 37 parallel cultures following exposure to MMS

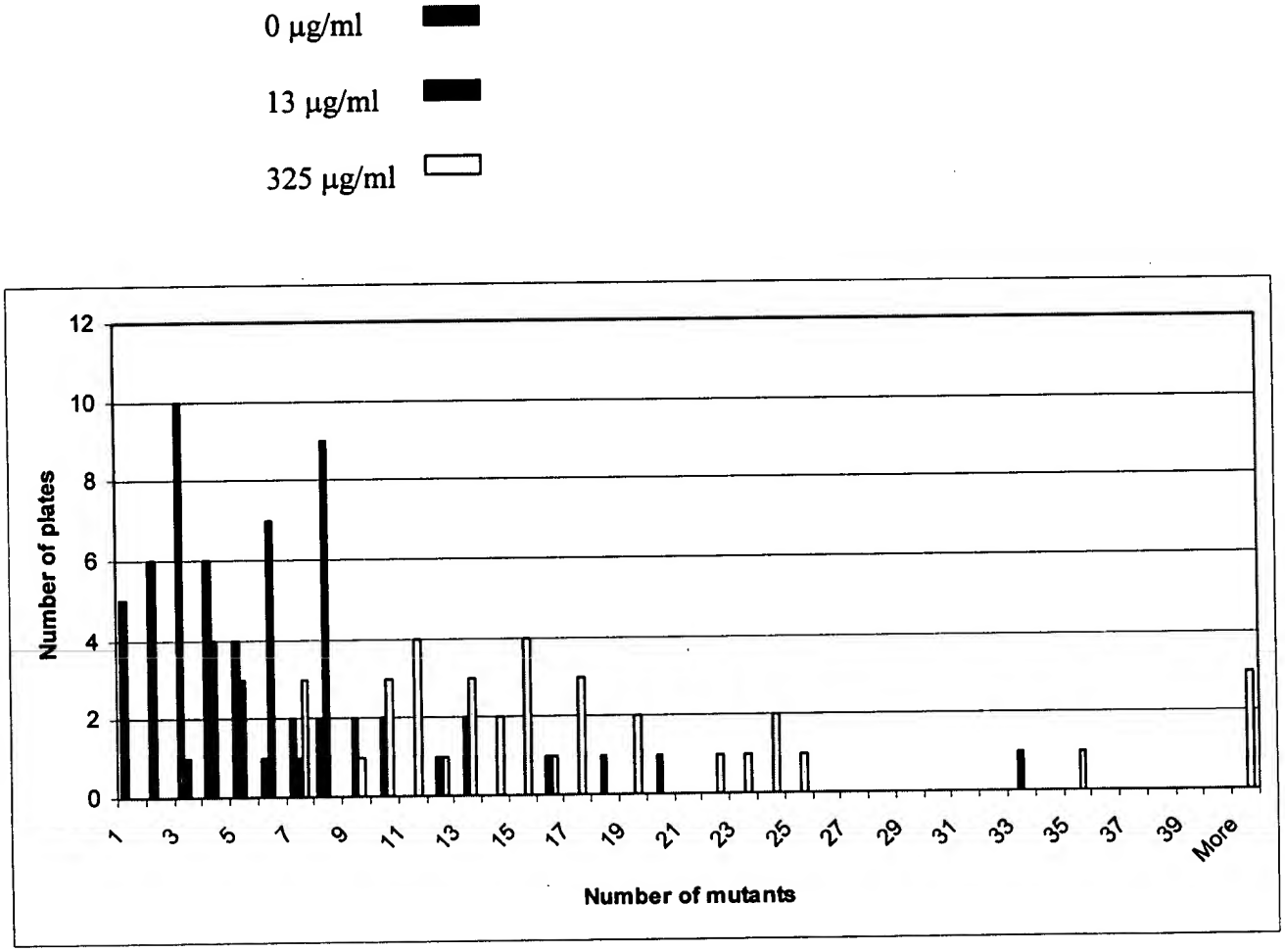
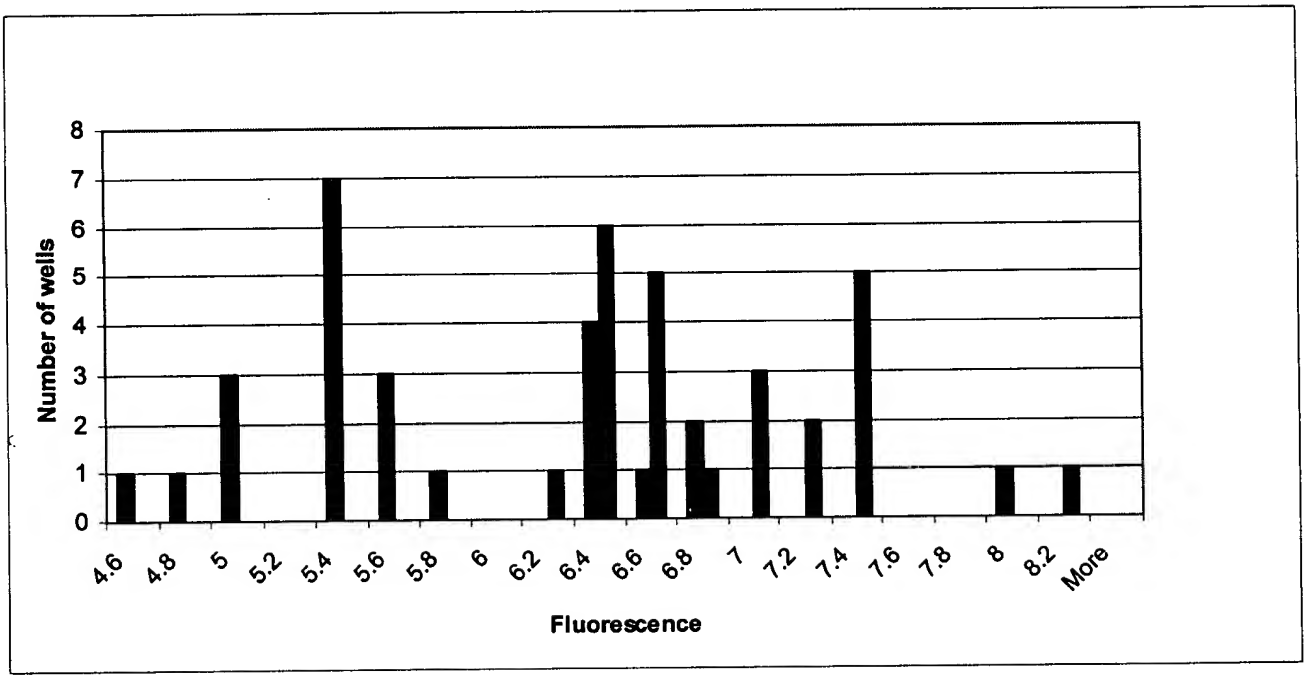


Figure 9 Histograms of mutant colony numbers and fluorescence emission from 24 parallel cultures following exposure to MNNG

0 μg/ml 1.7 μg/ml

a.) Fluorescence emission values



b.) Revertant colony numbers

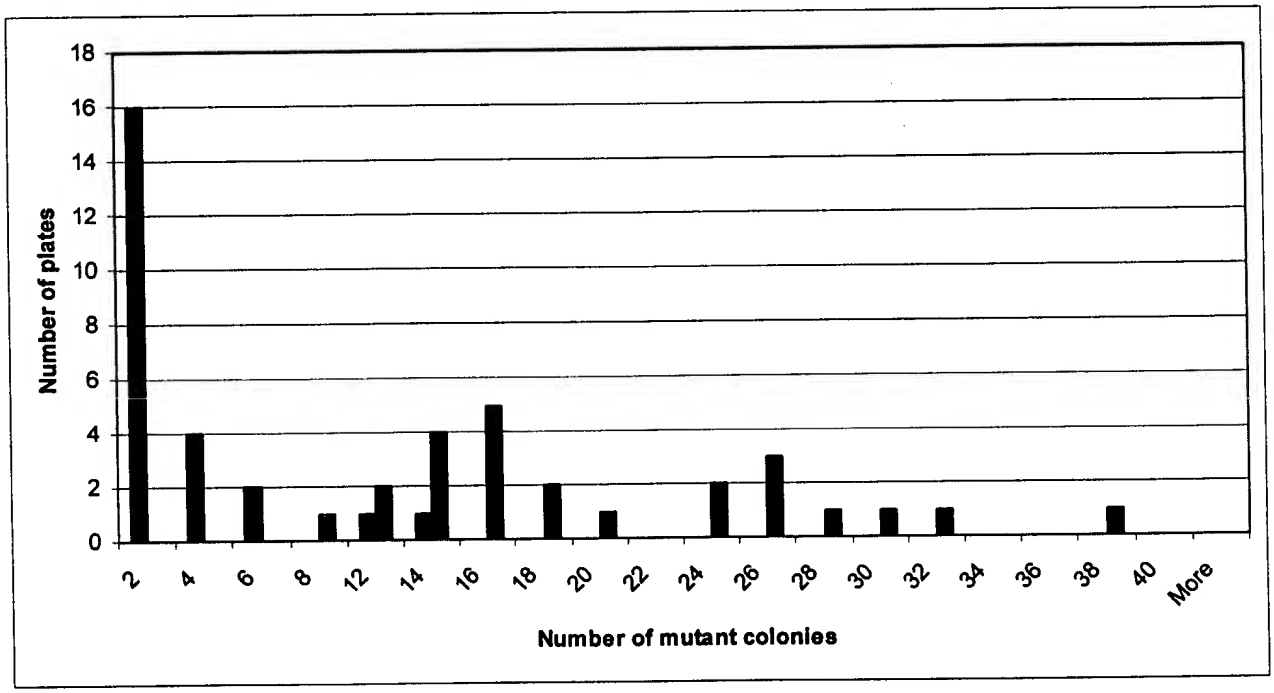


Figure 10 Histogram of fluorescence emission from 84 parallel cultures following exposure to MMS

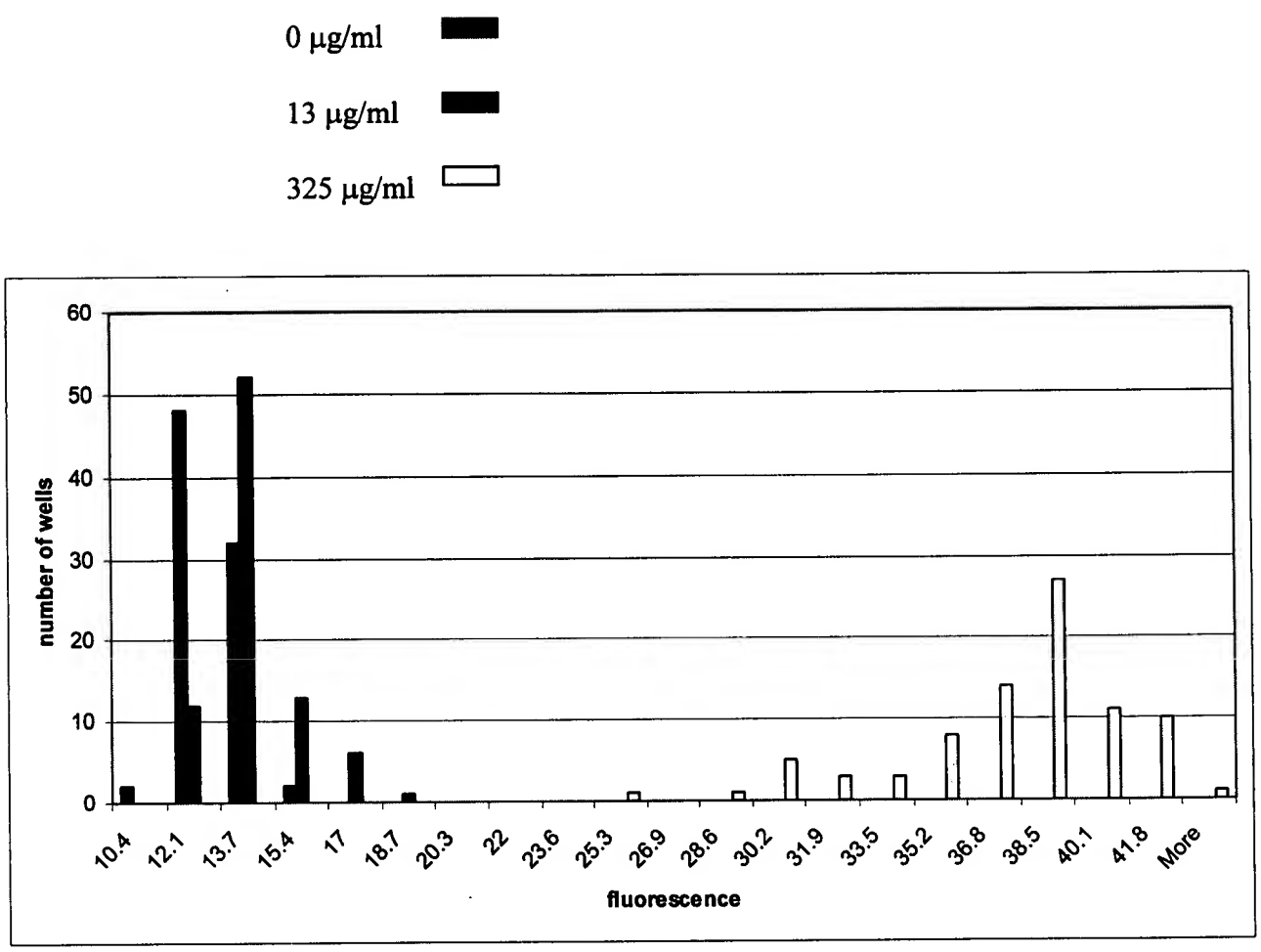


Figure 11 Histogram of fluorescence emission from 84 parallel cultures following exposure to MNNG

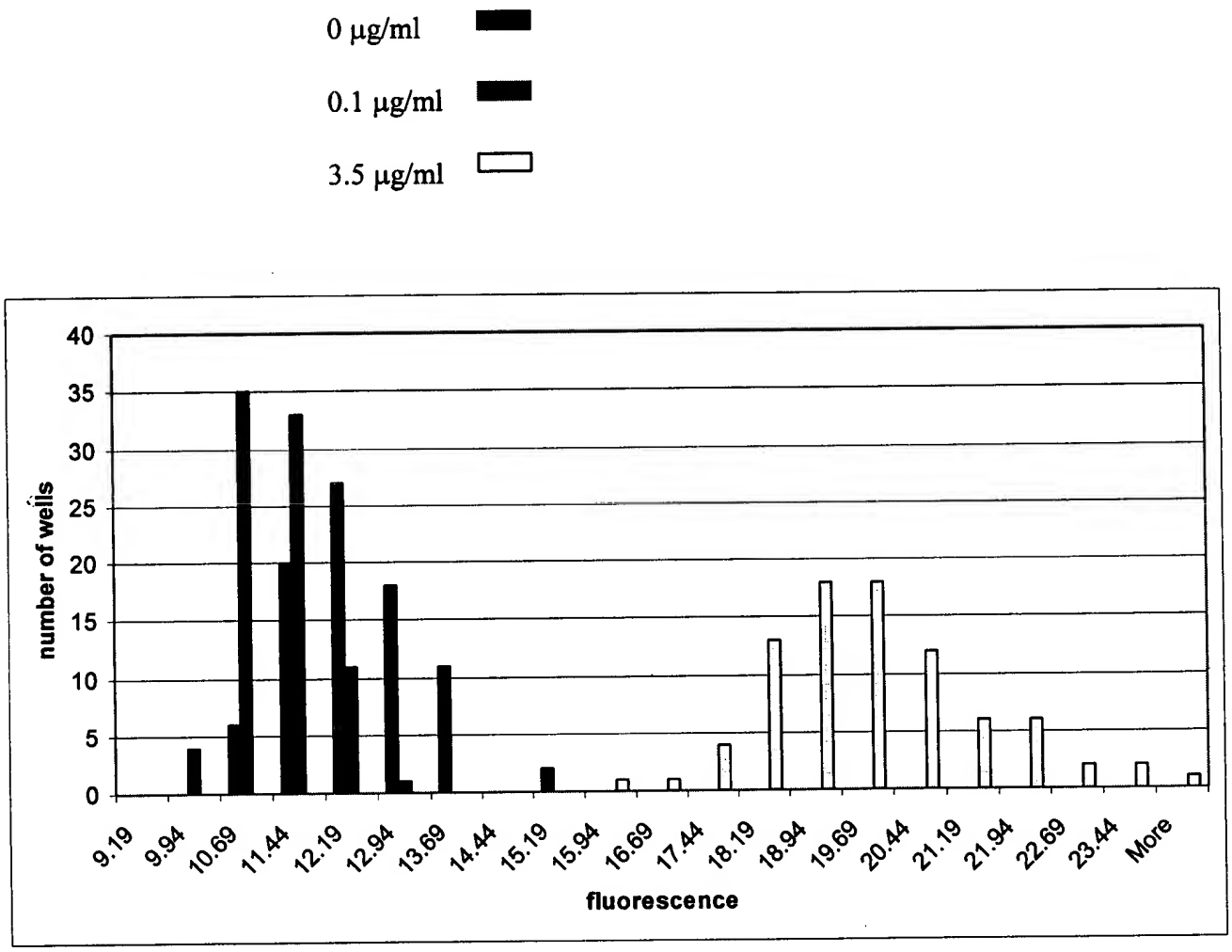


Figure 12 Histogram of fluorescence emission from 84 parallel cultures following exposure to 254nm UVC

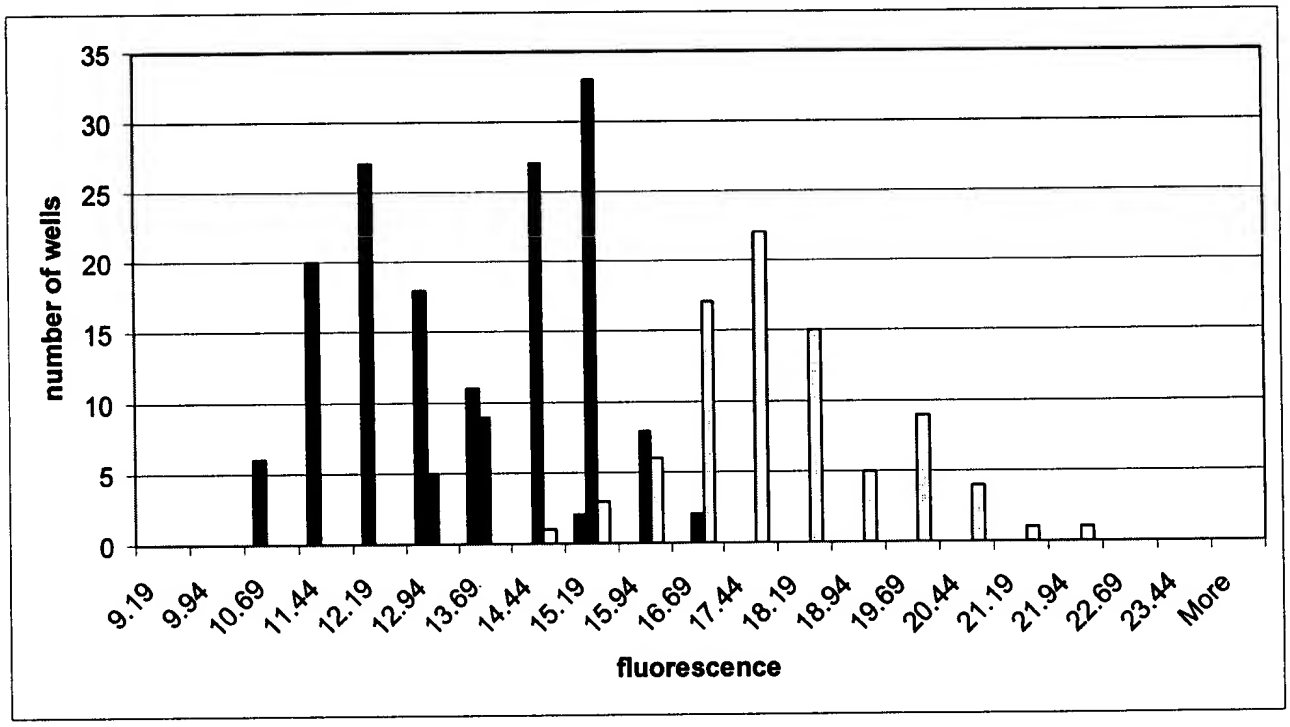


Figure 13

GenBank ACCESSION M13387

umuD protein

MLFIKPADLREIVTFPLFSDLVQCGFPSPAADYVEQRIDLNQLL

IQHPSATYFVKASGDSMIDGGISDGDLLIVDSAITASHGDIVIAAVDGEFTVKKLQLR
PTVQLIPMNSAYSPITISSEDTLDVFGVVIHVVKAMR"

umuC protein

MFALCDVNAFYASCETVFRPDLWGKPVVVLSNNDGCVIARNAEA

KALGVKMGDPWFKQKDLFRRCGVVCFSSNYELYADMSNRVMSTLEELSPRVEIYSI
DE

AFCDLTGVRNCRDLTDFGREIRATVLQRTHLTVGVGIAQTKTLAKLANHAAKKWQR
QT

GGVVDLSNLERQRKLMSALPVDDVWGIGRRISKKLDAMGIKTVLDLADTDIRFIRKH
F

NVVLERTVRELRGEPCLQLEEFAPTKQEIICSRSFGERITDYPSMRQAICSYAARAAE

KLRSEHQYCRFISTFIKTSPFALNEPYYGNSASVKLLTPTQDSRDIINAATRSLDAIW

QAGHRYQKAGVMLGDFFSQGVAQLNLFDDNAPRPGSEQLMTVMDTLNAKEGRGT
LYFA
GQGIQQQWQMKRAMLSPRYTTRSSDLLRVK

The Gene

1 aaaatcagca gcctatgcag cgacaaatat tgatagcctg aatcagtatt gatctgctgg
61 caagaacaga ctactgtata taaaaacagt ataacttcag gcagattatt atgttgttta
121 tcaagcctgc ggatctccgc gaaattgtga cttttccgct atttagcgat cttgttcagt
181 gtggctttcc ttcaccggca gcagattacg ttgaacagcg catcgatctg aatcaactgt
241 tgatccagca tcccagcgcg acttacttcg tcaaagcaag tggtgattct atgattgatg
301 gtggaattag tgacggtgat ttactgattg tcgatagcgc tattaccgcc agccatggtg
361 atattgtcat cgctgctgtt gacggcgagt ttacggtgaa aaaattgcaa ctacgcccga
421 cggtacagct tattcccatg aacagcgcgt actcgcccat taccatcagt agtgaagata
481 cgctggatgt ctttggtgtg gtgatccacg tcgttaaggc gatgcgctga tgtttgccct
541 ctgtgatgta aacgcgtttt atgccagctg tgagacggtg tttcgccctg atttatgggg
601 taaaccggtg gttgtgctat cgaataatga cggttgcgtt atcgcccgaa acgctgaggc

661 aaaggcgctt ggcgttaaaa tgggcgatcc ctggttcaaa caaaaagatc tgtttcgtcg
721 ctgtggcgtg gtttgcttta gcagcaatta tgagctttac gcagacatga gcaatcgggt
781 gatgtcgacg ctggaagagc tatcgccccg cgtcgagatt tacagtattg atgaggcatt
841 ctgcgatctg acaggtgtgc gtaattgtcg cgatctgact gattttggca gagaaattcg
901 cgcaacggtg ctacaacgta cccatcttac tgttggtgtg gggatcgccc agaccaaaac
961 gctggctaag cttgccaatc atgcggcaaa aaaatggcag cggcagacgg gtggggtggt
1021 ggatttatca aatctggaac gccagcgtaa attaatgtct gctctccccg tggatgacgt
1081 ctgggggatt ggacggcgga tcagcaaaaa actggacgcg atggggatca aaaccgttct
1141 cgatttggcg gatacagata tccggtttat ccgtaaacat tttaatgtcg tgctcgaaag
1201 aacggtgcgt gaactgcgcg gcgaaccctg tttgcaactg gaagagtttg caccgacgaa
1261 gcaggaaatt atctgttccc gctcgtttgg tgaacgcatc acggattatc cgtcgatgcg
1321 gcaggccatt tgtagttacg ctgcccgggc ggcggaaaaa cttcgcagcg agcatcaata
1381 ttgtcggttt atctccacgt ttattaagac gtcaccattt gcgctcaatg aaccttatta
1441 cggcaatagc gcgtcggtaa aactgctgac gcccactcag gacagcaggg atatcattaa
1501 cgctgctacg cgatctctgg atgccatctg gcaagcgggc catcgttacc aaaaagcggg
1561 cgtgatgctg ggggatttct tcagtcaggg agtcgcgcag ctcaatttat tcgatgacaa
1621 cgcaccgcgc cccgggagtg agcaattgat gacggtaatg gatacactga atgctaaaga
1681 gggcagagga acactctatt ttgccgggca ggggatccag caacaatggc agatgaagcg
1741 agccatgctt tcaccacgtt atacaacgcg aagttctgat ttactgaggg tcaaataaat
1801 atagcggcag gaaaaaa

Fig 13, cont'd

Figure 14

gfp mut2

protein

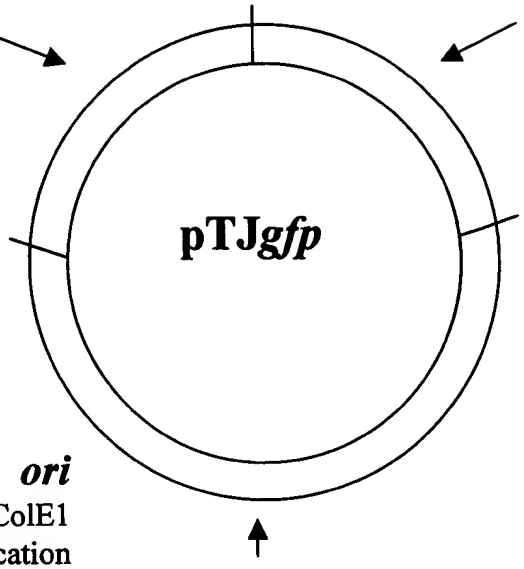
MSKGEELFTGVVPILVELDGDVNGHKFSVSGEGEGDATYGKLTLKFICTTGKLPVPW
PTLVTTFAYGLQCFARYPDHMKQHDFFKSAMPEGYVQERTIFFKDDGNYKTRAEVK
FEGDTLVNRIELKGIDFKEDGNILGHKLEYNYNSHNVYIMADKQKNGIKVNFKIRHNI
EDGSVQLADHYQQNTPIGDGPVLLPDNHYLSTQSALSKDPNEKRDHMVLLEFVTAA
GITHGMDELYK

gene

1 aagctttatt aaaatgtcta aggtgaaga attattcact ggtgttgtcc caattttggt
61 tgaattagat ggtgatgtta atggtcacaa attttctgtc tccggtgaag gtgaaggtga
121 tgctacttac ggtaaattga ccttaaaatt tatttgtact actggtaaat tgccagttcc
181 atggccaacc ttagtcacta ctttcgcgta tggtcttcaa tgttttgcta gatacccaga
241 tcatatgaaa caacatgact ttttcaagtc tgccatgcca gaaggttatg ttcaagaaag
301 aactattttt ttcaaagatg acggtaacta caagaccaga gctgaagtca agtttgaagg
361 tgatacctta gttaatagaa tcgaattaaa aggtattgat tttaaagaag atggtaacat
421 tttaggtcac aaattggaat acaactataa ctctcacaat gtttacatca tggctgacaa
481 acaaaagaat ggtatcaaag ttaacttcaa aattagacac aacattgaag atggttctgt
541 tcaattagct gaccattatc aacaaaatac tccaattggt gatggtccag tcttgttacc
601 agacaaccat tacttatcca ctcaatctgc cttatccaaa gatccaaacg aaaagagaga
661 ccacatggtc ttgttagaat ttgttactgc tgctggtatt acccatggta tggatgaatt
721 gtacaaataa ctgcag

Figure 15

The structure and sequence of the construct



***umu*DC**
bp 1-968
i.e. all of *umuD* + *umuC* to bp968 as described by Perry et al.(1985)

gfp
740bp fragment, including rbs, PCR amplified from GFPmut2 described by Cormack et al., (1996) (mutated from the wildtype at S65A, V68L, S72A) using the method of Matthysse et al (1996)

Bla
β-lactamase, a selectable marker

ori
(a ColE1 replication origin)

pSE117
7kb HindIII-EcoRI fragment
(i.e. excluding *umuC* from bp 969 onwards)
as described by Marsh and Walker (1985),
carrying both the ColE1 replication origin
and the antibiotic resistance selectable marker